Evaluation of the Pear Ester Kairomone as a Formulation Additive for the Granulovirus of Codling Moth (Lepidoptera: Tortricidae) in Pome Fruit

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ABSTRACT (*E,Z*)-2,4-decadienoate (pear ester) is a larval kairomone for the codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae). Orchard studies were conducted in 2005 and 2006 in apple, *Malus domestica* Borkhausen, and pear, *Pyrus communis* L., to evaluate a 5% active ingredient (AI), microencapsulated formulation of pear ester (PE-MEC) as an insecticidal additive for the codling moth granulovirus (CpGV). Although CpGV applied at 5–15-d intervals at commercial rates (2.2×10^{12}–10^{13} granules per ha) killed the majority (82–94%) of larvae found inside infested fruit, it did not eliminate significant damage, i.e., 30–92% fruit injury at harvest versus 51–82% in controls. PE-MEC treatments had significant but inconsistent results in our tests. In apple (mixed cultivars), PE-MEC (3.7–4.7 g [AI]/ha) plus CpGV reduced the percentage of fruit injured during the second but not the first larval generation, compared with CpGV alone, but there no was no additional population reduction (live larvae collected from infested fruit and tree bands). In 'Bartlett' pear, PE-MEC (3.7 g [AI]/ha) plus CpGV significantly increased larval mortality and reduced deep fruit entries at harvest over CpGV alone in 2006, but similar improvements were not observed in 2005 when a lower rate (1.5 g [AI]/ha) was tested. Surprisingly, compared with untreated controls, the PE-MEC formulation alone also reduced fruit injury (mid-season in Bartlett) and larval survivorship inside infested fruit at harvest (2006 apple tests and both years in Bartlett). Although pear ester seems amenable as a kairomonal adjuvant for use with insecticides, our inconsistent data with CpGV in apple and pear suggest practical improvements in formulation and application strategies (e.g., to optimize and maintain attractive release rates) are needed.

KEY WORDS kairomone, CpGV, ethyl (*E,Z*)-2,4-decadienoate, apple, pear

The granulovirus of the codling moth, *Cydia pomonella* (L.) (Lepidoptera: Tortricidae), CpGV, is highly virulent and specific to codling moth and closely related species (Gröner 1986, Huber 1986). Numerous orchard trials have demonstrated the potential of CpGV to control codling moth populations when applied as aqueous sprays to coincide with hatching of neonate larvae that ingest virus granules before or during entry into fruit (Huber 1986, Falcon and Huber 1991, Cross et al. 1999, Lacey et al. 2007). CpGV has been used by European orchardists since 1988 (Cross et al. 1999), and more recently, in North America (Lacey et al. 2004b).

Although CpGV is effective at controlling codling moth, a major disadvantage of the virus is its sensitivity to abiotic factors, particularly the damaging UV-B portion of sunlight (280–320 nm). Reapplication is typically required at 7–10-d intervals during periods of egg hatch (Glen and Clark 1985, Huber 1986, Jaques et al. 1987, Kienzle et al. 2003, Arthurs et al. 2005). An additional drawback of the virus is that the slow rate of kill allows the occurrence of shallow entries or "stings" in sprayed fruit. The short residual activity and the need to cull fruit damaged by the treated larval generation increases the cost of CpGV (materials and labor), limiting commercial adoption.

A variety of strategies to improve the larvicidal activity of CpGV has been investigated. The application of increased concentrations of virus or more frequent applications of lower concentrations have been tried with limited success, and these actions did not eliminate economic damage to fruit (Glen and Payne 1984, Dickler and Huber 1986, Ballard et al. 2000a, Arthurs et al. 2005). Formulation additives have been investigated with the goal of increasing the uptake, persistence, or both of CpGV in the field. Substances including molasses, sucrose, fructose, and sorbitol have been reported to enhance feeding and CpGV uptake slightly, although the rates used are considered high for routine field use (Ballard et al. 2000a). The UV screen oxybenzone and skimmed milk, which also may function as a feeding stimulant, have reportedly improved the persistence of CpGV slightly through in-

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creased solar protection (Krieg et al. 1980; Keller 1973, Charmillot et al. 1998). The use of several adjuvants, including the spreader/sticker NuFilm-17 and carnauba wax-based sunburn protectant Raynox did not extend the persistence of CpGV formulations in orchard trials (Lacey et al. 2004a) or laboratory tests (Arthurs et al. 2006). However, the incorporation of virus into a water-soluble, lignin-based carrier through spray drying did significantly extend the persistence of virus in laboratory tests with a solar simulator (Arthurs et al. 2006).

The pear ester, ethyl (*E,Z*)-2,4-decadienoate, a key odorant of ripe pears, has recently been identified as a kairomonal attractant for both sexes of adult codling moth (Light et al. 2001) and neonate larvae (Knight and Light 2001). The possibility of combining pear ester with insecticides as attract-and-kill formulations in paste, bait stations or sprayable formulations has been noted (Knight and Light 2001, Knight et al. 2002). Pear ester can stimulate oviposition by codling moth (Knight and Light 2004a) and has been reported to influence the pattern of egg deposition in both apple and pear (Pasqualini et al. 2005a). The authors in this latter study reported that eggs, on average, were laid further from fruit on trees treated with a black gel formulation of pear ester. The potential use of kairomones to cause host disruption during the susceptible period of neonate wandering has been suggested as a promising avenue to enhance control of codling moth (Hughes et al. 2003).

Ballard et al. (2000a) reported orchard plots treated with CpGV formulated with 0.08% (*E,E*)-α-farnesene, 10% molasses, or 10% sorbitol had reduced levels of fruit with deep entries compared with the application of CpGV alone. Extracts of apple flesh and skin [but not (*E,E*)-α-farnesene alone] also gave significant reductions in the lethal exposure time for neonate larvae placed on leaf discs treated with CpGV at 10^7 occlusion bodies per ml (Ballard et al. 2000a). By contrast, pear ester is more chemically stable than (*E,E*)-α-farnesene (Light et al. 2001), and it is attractive to codling moth neonates at concentrations 10- to 1,000-fold lower than to (*E,E*)-α-farnesene (Knight and Light 2001). Light (2007) significantly reduced levels of nut injury in walnut (*Juglans* spp.) by adding a microencapsulated formulation of pear ester to low rates of several insecticides, including CpGV. In this article, we report on four orchard studies in pome fruit evaluating pear ester as an insecticidal synergist for CpGV in different cultivars of apple and 'Bartlett' pear.

Materials and Methods

Source of Virus and Pear Ester. Commercial preparations of CpGV 'Cyd-X' (Certis USA, Columbia, MD) and Carpovirusine (Arysta LifeScience, Cary, NC) containing 3×10^{13} and 10^{13} granules/liter, respectively, were used in all tests. Pear ester was a 5% active ingredient (AI) microencapsulated formulation (PE-MEC, Trécé Inc., Salinas, CA).

Orchard Treatments. Codling moth flight activity in test blocks was always monitored with delta-shaped traps baited with sex pheromone (codlemone), and, with the exception of the 2005 apple study, pear ester lures (separate traps) placed in the upper canopy. Lures and sticky trap liners were changed every 2–3 wk. A minimum of two traps were placed per orchard, and they were checked every 1–2 d at the start of the season to determine the start of sustained flight of male moths (biofix). Timing of treatments was determined using the date for biofix and a degree-day (DD) phenology model for codling moth (Brunner et al. 1987). Treatments were applied as dilute sprays during the main period of egg hatch in the orchard. CpGV was kept refrigerated and mixed the morning of use to ensure viability. Spray applications were made during calm wind conditions ($\leq$0.5 m/s). A one- to two-tree buffer row was used between adjacent plots to minimize overspray or spray drift (a flexible tarpaulin screen held by 2–4 assistants was also used in apple tests). No additional insecticides were applied to test plots during tests. All orchards were supplied with under-tree sprinklers, which minimized pesticide washoff during irrigation. Specific details for each study are presented below.

2005 Apple Study. The study orchard was a 0.5-ha block of 15-yr-old 'Golden Delicious' (Smoothee strain on EMLA 7 rootstock) planted at a spacing of 3.7 by 5.5 m and located at the USDA–ARS field station near Moxee, WA. Experimental treatments were applied using a motorized backpack airblast sprayer (10-liter capacity, model SR 420, Stihl, Virginia Beach, VA). Treatments applied were Cyd-X alone (23.4 ml/100 liters, equivalent to 6.6×10^{12} granules per ha), Cyd-X (same rate) plus PE-MEC (7.81 ml formulated material/100 liters, equivalent to 3.7 g [AI]/ha), PE-MEC alone (same rate), and untreated controls (sprayed with water and NuFilm17). The second generation was treated using the same plots, but with reduced rates of CpGV (2.2×10^{12} granules/ha). Sprays were directed around the trees to provide complete coverage of foliage and fruit at 935 liter/ha. The spreader-sticker NuFilm-17 (Miller Chemicals and Fertilizer Corp., Hanover, PA) was included in all treatments at 46.8 ml/100 liter. The sprayer was frequently agitated to prevent settling and rinsed between treatments.

Experimental Design. The study was a complete randomized block, with 10 single tree replicates (eight for untreated controls and PE-MEC alone). Initial applications were made on 26 May (226 DD when the model's biofix predicted 2% egg hatch), and treatments were reapplied every 7 d (334, 389, 458, 564, 677, and 795 DD) for a total of seven applications against the first larval generation. Applications against the second generation started 28 July (1,205 DD) for a total of six weekly applications (1354, 1516, 1655, 1786, and 1,901 DD).

Field Assessments. Codling moth fruit injury was assessed in situ from a random (blind pick) sample of 50 fruit on all sprayed trees at the end of the first and second larval generations: between 20 and 22 July

(1034–1075 DD) and 12–15 September (2022–2054 DD), respectively. On each occasion, subsamples of infested fruit (50 and 30 fruit on each treated or water control tree, respectively) were picked, returned to the laboratory, destructively sampled, and examined at 10× magnification to quantify larval survival. The proportion of "shallow stung" fruit (i.e., ≤5 mm from the surface) also was noted as proxy for virus dosage consumed and speed of kill (Arthurs et al. 2005). Exit holes caused by mature larvae leaving fruit were included as live larvae. Fruit were maintained at 12°C for up to 3 wk before processing. Corrugated cardboard bands were used to assess the number of overwintering mature larvae in each plot (Arthurs et al. 2005). Bands were stapled around the base of treated and control trees when diapause-destined larvae started exiting unsprayed fruit on 24 August (1,771 DD) and examined for cocooned larvae on 3 October (2,198 DD). Bands were periodically renewed due to bird damage. Conditions throughout the study were predominantly sunny and dry with only 0.89 cm of rainfall. Average maximum and minimum daily temperatures during the study were 26.7°C (range, 17.9–37.0) and 9.2°C (range, 2.0-16.8) during the first larval generation and 29.2°C (range, 16.4–37) and 10.8°C (range, 5.4–16) during the second larval generation.

2006 Apple Study. The study plot was a 0.6-ha mixed cultivar portion of an 11-yr-old 1.4-ha orchard located at the USDA–ARS field station. Trees were planted at a spacing of 2.4 by 4.9 m. Each replicate block consisted of four trees, 'Golden Delicious' (Mullens strain on EMLA 7 rootstock), 'Red Fuji' (BC#2 on EMLA 26 rootstock), 'Scarlet Gala' (EMLA 26 rootstock), and 'Nured Spur Red Delicious' (EMLA 7 rootstock). The planting sequence was always Golden Delicious, Red Fuji, Scarlet Gala, and Nured Spur Red (from north to south) with a one tree open spacing between blocks. Sprays were applied with a diesel powered air-blast sprayer (Hauff Company, Yakima WA) pulled with an all terrain vehicle. The sprayer (95-liter capacity) was calibrated to apply 935 liters/ha operating at 1,034 kPa and 2.4-kph forward speed. Treatments included Cyd-X alone (23.4 ml/100 liter, 6.6×10^{12} granules per ha), Cyd-X plus PE-MEC (10 ml/100 liters, 4.7 g [AI]/ha), PE-MEC alone (same rate), and untreated controls. No spreader, stickers, or both were used.

Experimental Design. Experimental treatments were applied to individual four-tree blocks with five replicates per spray treatment assigned within individual rows of the orchard. The study was a two-way factorial, with spray program and cultivar as the tested variables, arranged within a randomized complete block design. Initial applications were made 1 June, with treatments reapplied at 5–15-d intervals for a total of five applications against the first generation (288, 348, 460, 605, and 944 DD). The second larval generation was not treated.

Field Assessments. Codling moth fruit injury was assessed in situ at the end of the first larval generation on 18–19 July (1,047–1,065 DD). In general, all fruit on test trees were inspected; although, where fruit load was heavy (notably on Golden Delicious), a subsample made up of two of the main scaffold branches (on opposite sides on each tree) was selected for evaluation. Subsamples of infested fruit (up to 20 per tree. depending on availability) were sampled to quantify larval survivorship and shallow stings, as for 2005. Conditions were predominantly sunny and dry with only 2.18 cm of rainfall throughout the study. Average maximum and minimum daily temperatures during the study were 27.4°C (range, 16.8–37.0) and 10.6°C (range, 5.4–19.4).

2005 Pear Study. The study orchard was a 45-yr-old Bartlett block (0.4 ha) planted at a spacing of 3.7 by 7.3 m and located at the Southern Oregon Research and Extension Center (SOREC), Medford, OR. Treatments were applied with a tractor-driven airblast sprayer (Rears Mfg., Eugene, OR) fitted with dilute nozzles and calibrated to apply 1,262 liters/ha operating at 862 kPa with 2.4-kph forward speed. Treatments were Carpovirusine (78 ml/100 liter, 10^{13} granules per ha), Carpovirusine plus PE-MEC (2.3 ml of formulated material per 100 liters, 1.5 g [AI]/ha), PE-MEC alone (same rate) and untreated controls. No spreaders, stickers, or both were used.

Experimental Design. The study was a complete randomized block with four single tree replicates. Initial applications were made on 21 May, and treatments reapplied at 12–17-d intervals for a total of six applications until harvest (208, 358, 486, 658, 995, and 1,289 DD).

Field Assessments. Codling moth injury was assessed on two occasions: 22 June, mid-season after three applications (558 DD), and 28 July, preharvest shortly after the final application (1,316 DD). On each occasion, 50 fruit were picked and evaluated in the laboratory. Fruit was also sampled near harvest on 8 August (1,595 DD); 100 fruit were picked, and those infested were dissected in the laboratory, to quantify shallow stings (≤5 mm) and larval mortality. Average maximum and minimum daily temperatures during the study were 28.4°C (range, 13.5–39.1) and 11.2°C (range, 3.0–18.1). Weather conditions were generally dry through the treatment period with the exception of precipitation during the first generation in late May and early June: 1.7 cm between the first and second applications and 1.0 cm between the second and third applications, after which precipitation was always less that 0.2 cm between applications.

2006 Pear Study. The study orchard was a 10-yr-old mixed cultivar pear orchard planted at a spacing of 2.8 by 4.5 m and located at SOREC. The study was conducted in Bartlett (other cultivars were present but not sampled). Treatments were applied with an airblast sprayer (Rears Mfg.) pulled by a Rex tractor. The sprayer was fitted with concentrate nozzles and calibrated to deliver 748 liter/ha operating at 862 kPa and 2.4 kph. Treatments included Cyd-X alone (29.3 ml/100 liter, equivalent to 6.6×10^{12} granules per ha), Cyd-X plus PE-MEC (10 ml/100 liter, 3.7 g [AI]), PE-MEC alone (same rate), and untreated controls. No spreaders or stickers were used.

Experimental Design. The study was a randomized block design with five replicates (10–20 trees per

Table 1. Orchard evaluation in Golden Delicious of CpGV (Cyd-X formulation) applied with and without pear ester (PE-MEC) against codling moth larvae, Moxee, WA, 2005

Treatment	First generation[a]			Second generation[a]			
	% fruit injury[b]	% shallow stings	% mortality	% fruit injury[b]	% shallow stings	% mortality	Tree bands[c]
Untreated	18.0 ± 2.2ab	27.0 ± 3.4b	35.4 ± 4.7b	58.8 ± 2.3a	23.9 ± 2.6b	22.5 ± 1.8b	95.3 ± 4.5a
PE-MEC	19.0 ± 0.9a	28.8 ± 1.1b	41.1 ± 1.5b	53.1 ± 2.8ab	26.3 ± 4.4b	24.3 ± 3.0b	99.3 ± 7.3a
CpGV	11.5 ± 1.7bc	83.1 ± 2.4a	93.5 ± 1.6a	48.0 ± 3.1b	59.0 ± 1.0a	82.1 ± 1.6a	33.4 ± 3.0b
CpGV + PE-MEC	11.1 ± 3.2c	85.3 ± 2.9a	93.9 ± 1.3a	39.5 ± 2.5c	55.1 ± 2.0a	77.1 ± 1.4a	34.4 ± 3.8b

Column means ± SEM (eight to 10 single-tree plots) followed by different letters were significantly different ($P < 0.05$; Fisher LSD).
[a] Evaluations were made following seven weekly applications of CpGV (6.6×10^{12} granules per ha) and PE-MEC (3.7 g [AI]/ha) against the first larval generation and a further six weekly CpGV applications (2.2×10^{12} granules per ha) against the second larval generation.
[b] Data show fruit injury (n = 400–500) and shallow entries (≤5 mm) and larval mortality (n = 240–500 infested fruit).
[c] Number of diapause-destined larvae captured per tree, includes live larvae removed during fruit evaluations.

replicate). Initial applications were made 18 May and treatments generally reapplied at 14-d intervals (range, 6–14) for a total of seven applications until harvest (309, 438, 527, 712, 1,053, 1,331, and 1,669 DD).

Field Assessments. Bartlett fruit were sampled in situ on 7 July, mid-season after five applications (1,084 DD), and again on 15 August at harvest (1,950 DD). Bartlett pears were picked on the harvest assessment and dissected in the laboratory to quantify larval mortality and shallow entries (≤5 mm). One hundred fruit were examined per replicate. Average maximum and minimum daily temperatures during the study were 28.6°C (range, 13.6–40.6) and 12.3°C (range, 3.8-20.7). Weather conditions were generally dry throughout the treatment period with the exception of precipitation during late May and early June: 1.5 cm between the first and second applications, 0.6 cm between the second and third applications, 1.9 cm between the third and fourth applications, and then 0.4 cm the week before harvest.

Data Analysis. Treatment effects in all studies were compared using one- and two-way univariate analysis of variance (ANOVA) (SPSS Inc. 2003). Significant F-ratio means were further separated with Fisher least significant difference (LSD) multiple comparison test, at $P < 0.05$. Where necessary, the arcsine and log $(n + 1)$ transformation were used to normalize proportion and count data respectively, before analysis.

Results

2005 Apple Study. Fruit injury in CpGV treatments was lower than the untreated and PE-MEC treatments after the first larval generation, although not all comparisons were significant at $P < 0.05$ (Table 1). Larval mortality in CpGV-treated fruit was >93%, with most neonates dying near the surface of fruit in the first stadium, as reflected in the high percentage of shallow stings. In both the untreated control and PE-MEC treatment, the majority of larvae reached the core to feed on the seeds. There were no significant effects of applying PE-MEC, either alone or in combination with CpGV, during the first generation.

Cumulative moth catches increased from the first to the second generation in this orchard (i.e., from an average of 87–125 males per pheromone trap), and levels of fruit injury in the untreated plots also increased, i.e., from 18 to 59% (Table 1). Fruit injury in the second generation was again significantly lower in both CpGV treatments versus the untreated and PE-MEC treatments, but now also lower in the CpGV + PE-MEC versus CpGV treatment. Larval mortality inside infested fruit treated with the reduced rate of CpGV was lower (77–82%) with fewer shallow stings in fruit (55–59%), compared with the first generation, although still higher than in the untreated control and PE-MEC treatments. Larval mortality inside CpGV + PE-MEC-treated fruit was lower compared with CpGV alone (although not significantly lower). Overall, effects on overwintering codling moth populations (i.e., surviving experimental treatments) are unclear. Assessments of tree bands showed 64–65% fewer cocooning larvae in CpGV-treated trees, and no differences due to the PE-MEC treatment. Due to the experimental design, these tree bands may have been contaminated with larvae migrating from nearby unsprayed trees.

2006 Apple Study. The population density of codling moth was extremely high in this apple block with an average cumulative moth catch of 314 per pheromone trap during the first flight. Levels of fruit injury did not differ among spray treatments ($F_{3,59} = 0.4$; $P = 0.75$); however, a significant cultivar effect was detected ($F_{3,\ 59} = 14.5$; $P < 0.0001$). There was less injury on Red Fuji compared with other cutivars; when Red Fuji was removed from the data set, cultivar was not a significant factor ($F_{2,\ 45} = 0.6$; $P = 0.53$). Because the interaction of spray treatment and cultivar was not significant ($F_{9,\ 59} = 0.5$; $P = 0.87$), data were pooled among all cultivars within each four-tree block for one-way analysis; however, no significant differences were found between treatments (Table 2).

Cultivar was not a significant explanatory factor for mortality of larvae inside infested fruit ($F_{3,\ 44} = 1.0$; $P = 0.41$), and there was no interaction with spray treatment ($F_{9,\ 44} = 1.5$; $P = 0.18$). Because larval mortality was not affected by cultivar and relatively few infested fruit occurred on some trees (i.e., <10), data for larval mortality and shallow entries also were pooled within each plot for analysis (Table 2). Percentage of larval mortality and shallow stings were not different for CpGV + PE-MEC versus CpGV alone, although both

Table 2. Orchard evaluation in mixed apple of CpGV (Cyd-X formulation) applied with and without pear ester (PE-MEC) against first generation codling moth larvae, Moxee, WA, 2006

Treatment[a]	% fruit injury[b]	% shallow stings	% mortality
Untreated control	37.3 ± 4.2 (1,432)[c]	22.6 ± 1.8a (245)	48.9 ± 4.3c
PE-MEC	32.8 ± 3.2 (1,800)	47.1 ± 4.8b (225)	65.0 ± 4.3b
CpGV	33.9 ± 6.6 (1,589)	67.7 ± 5.0c (198)	88.9 ± 4.0a
CpGV + PE-MEC	34.3 ± 4.0 (1,798)	66.0 ± 2.9c (224)	87.4 ± 3.4a

Column means ± SEM (five replicate four-tree blocks) followed by different letters were significantly different ($P < 0.05$; Fisher LSD).

[a] Evaluations made following five applications of CpGV (6.6×10^{12} granules per ha) and PE-MEC (4.7 g [AI]/ha) at 5–15-d intervals in tests against the first larval generation.

[b] Data show fruit injury on tree, with shallow entries (≤5 mm) and larval mortality evaluated from infested fruit (pooled among four cultivars; see Materials and Methods).

[c] Number fruit sampled in parentheses.

Table 4. Orchard evaluation in Bartlett pear of CpGV (Cyd-X formulation) applied with and without pear ester (PE-MEC) against codling moth larvae, Medford, OR, 2006

Treatment	Mid-season[a]	Harvest		
	% fruit injury[b]	% fruit injury	% shallow stings	% larval mortality
Untreated	13.0 ± 2.6a	50.6 ± 2.9a	32.7 ± 3.2d	48.9 ± 3.2d
PE-MEC	5.6 ± 1.7b	44.2 ± 3.3a	52.8 ± 5.2c	68.6 ± 2.7c
CpGV	6.2 ± 1.5b	30.0 ± 5.2b	72.6 ± 3.0b	88.8 ± 2.8b
CpGV + PE-MEC	6.0 ± 1.2b	27.0 ± 3.6b	89.2 ± 1.6a	94.6 ± 0.9a

Column means ± SEM (five plots, 10–20 trees per plot) followed by different letters were significantly different ($P < 0.05$; Fisher LSD).

[a] Evaluations were made after five or seven biweekly applications of CpGV (6.6 b$\times 10^{12}$ granules per ha) and PE-MEC (3.7 g [AI]/ha) on 7 July and 15 August, respectively.

[b] Data show fruit injury (n = 500 fruit); harvest data also shows shallow stings (≤5 mm) and larval mortality evaluated from infested fruit.

these treatments were significantly higher than controls and PE-MEC alone. Larval mortality and shallow stings were also significantly higher for PE-MEC alone versus the untreated control.

2005 Pear Study. Percentage of fruit injury was reduced in both CpGV treatments and PE-MEC alone both at mid-season and preharvest compared with the untreated control (Table 3). However, injury was not reduced in the CpGV + PE-MEC versus CpGV alone, in either case. Damage increased rapidly at harvest, due to second generation larvae, and no differences in fruit injury were observed among treatments at this time. The majority of damage in CpGV-treated fruit at harvest consisted of shallow entries, containing moribund or dead larvae, with no difference between CpGV and CpGV + PE-MEC. There were more dead larvae and shallow stings in PE-MEC alone compared with untreated controls at harvest.

2006 Pear Study. The percentage of fruit injury was reduced in both CpGV treatments at mid-season and harvest compared with the untreated control; injury also was reduced in PE-MEC alone at mid-season (Table 4). However, injury was not reduced in the CpGV + PE-MEC versus CpGV alone, in either case. As in 2005, harvest evaluations showed most neonates were killed before making deep entries in CpGV-treated fruit, although in this case both larval mortality and shallow stings were significantly higher in the CpGV + PE-MEC compare with CpGV alone. As in 2005, higher mortality and shallow stings were observed in the PE-MEC alone versus untreated controls.

Discussion

CpGV is a potent, but environmentally sensitive, microbial insecticide for codling moth. Field efficacy of CpGV is strongly impacted by temporal factors that determine the degree of exposure of larvae: time spent by larvae in host finding and penetration of fruit and the rapid loss of virus potency. It should be noted that because most trees in the test orchards remained unsprayed, our evaluations of codling moth damage do not reflect overall efficacy of CpGV treatments (which by controlling initial populations normally greatly reduce damage by the second larval generation), but rather they indicate the high pest population pressure used to compare treatments. The distribution of codling moth eggs on or near fruit or nuts (Jackson 1979) and their limited feeding before host penetration are the key biological factors that limit the effectiveness of CpGV. Conceptually, adjuvants that increase larval exposure to residues and ingestion of CpGV should improve the effectiveness of CpGV.

Several attributes of pear ester could make it an ideal adjuvant to improve the efficacy of CpGV. Pear ester is a strong attractant for codling moth neonates (Knight and Light 2001) and a stimulus for adult ovi-

Table 3. Orchard evaluation in Bartlett pear of CpGV (Carpovirusine formulation) applied with and without pear ester (PE-MEC) against codling moth larvae, Medford, OR, 2005

Treatment	Mid-season[a]	Preharvest	Harvest		
	% fruit injury[b]	% fruit injury	% fruit injury	% shallow stings	% larval mortality
Untreated	32.5 ± 2.8a	68.0 ± 5.7a	81.8 ± 8.0	11.5 ± 3.4c	24.8 ± 5.2c
PE-MEC	15.5 ± 3.6b	48.5 ± 4.6b	82.3 ± 3.5	25.0 ± 2.9b	50.6 ± 5.7b
CpGV	18.5 ± 5.3b	21.5 ± 2.8c	91.5 ± 1.3	61.3 ± 3.1a	85.7 ± 2.4a
CpGV + PE-MEC	12.0 ± 2.9b	16.5 ± 3.2c	81.3 ± 3.3	54.5 ± 4.2a	85.0 ± 2.8a

Column means ± SEM (four replicate single-tree blocks) followed by different letters were significantly different ($P < 0.05$; Fisher LSD).

[a] Evaluations were made after three (mid-season) or six biweekly applications of CpGV (10^{13} granules per ha) and PE-MEC (1.5 g [AI]/ha) on 22 June, 28 July, and 8 August, respectively.

[b] Data show fruit injury (n = 200–400 fruit); harvest data also show shallow stings (≤5 mm) and larval mortality evaluated from infested fruit.

position (Knight and Light 2004a). Spray applications of pear ester have extended the average distance that eggs are laid from pome fruit (Pasqualini et al. 2005a). Codling moth neonates may become infected with CpGV by walking or browsing on previously sprayed foliage in as little as 3.5 min, although the probability of dying from virus infection greatly increased with the time spent on the leaf surface (Ballard et al. 2000b). Before our study the potential of pear ester to increase the exposure of codling moth neonates to CpGV had been suggested but not well substantiated in field studies in pome fruit (Pasqualini et al. 2005b). In comparison, Light (2007) was able to further reduce injury in walnut by 47% with an eight-spray seasonal program of CpGV plus PE-MEC compared with CpGV alone.

The benefits of adding pear ester to CpGV in our field studies in pome fruit, however, were inconsistent and more limited: a moderate reduction of fruit injury in apple during the second but not the first generation; and in Bartlett pear, a moderate increase in larval mortality and the percentage of shallow versus deep stings at harvest in 2006, but no similar increase on other occasions. Several factors (discussed below) could contribute to these variable results seen among crops and seasons.

Factors affecting the responses of adult codling moth to pear ester have been studied (Light et al. 2001; De Cristofaro et al. 2004; Knight and Light 2005a,b). For example, the relative attractiveness of pear ester for adult codling moth varies between walnut and pome fruit (Light et al. 2001) and among apple and pear cultivars (Knight and Light 2004a, Knight et al. 2005). In Bartlett pear, pear ester lures loaded with higher rates (up to 40 mg [AI]) generally caught more males, suggesting dosage is also important (Knight and Light 2004b). Pear ester may become less attractive in late season, as releases of competing host volatiles increase. Maturing fruit releases higher quantities of aliphatic esters, alcohols, and some sesquiterpenes, such as (*E,E*)-alpha-farnesene, an attractant and ovipositional stimulant for adult codling moths (Wearing and Hutchins 1973, Sutherland et al. 1977) and neonate larvae (Landolt et al. 2000). Light et al. (2001) suggested that the decline in the number of males caught using pear ester lures later in the season in some apple and pear orchards was due to "olfactory masking" by natural sources of pear ester released from injured or ripening fruit or competing host volatiles. Unfortunately, the behavior of codling moth neonates to pear ester has not been well studied. Knight and Light (2001) found that codling moth neonates are attracted to extremely low rates of pear ester in filter paper bioassays, but they did not report the optimal doses eliciting these responses. Similar studies evaluating larval response to pear ester-treated host plant materials have not been conducted. Furthermore, the release rate of pear ester from the proprietary MEC formulation used in our tests has not been reported. This lack of information provides a serious limitation to our data interpretation on larval attractiveness. In the same way volatile release rates from lures were carefully developed to elicit optimal response for adult moths (Light et al. 2001, Knight and Light 2005a), neonates may respond to very specific quantities of pear ester (Knight and Light 2001). Although very low application rates may fall below a response threshold, higher application rates may cause rapid habituation and be detrimental for behavior modification. For example, a low density of evenly distributed capsules may form mini point sources of attraction, whereas higher densities may overwhelm or confuse larvae, resulting in them resorting to secondary volatile or other host finding cues (D. M. Light, personal communication). The interaction of capsule density, distribution on leaf surfaces and volume application rate is also likely important (Light, unpublished data). Such interactions on neonate behavior require further study; unfortunately, these factors as well as the dosage of CpGV applied and crop type were not standardized in our tests, making comparisons difficult.

Interestingly, the PE-MEC formulation alone generated significant reductions in both fruit injury (midseason in Bartlett pear) and larval survivorship (2006 tests with apple and both years in Bartlett pear). In comparison, no reductions in injury in walnut plots (Light 2007) or increased larval mortality (A.L.K., unpublished data) were previously found from the PE-MEC or a blank MEC formulation. An explanation for these latter effects is unclear but could be attributed to energy depletion in larvae eclosing from eggs deposited further from fruit (Pasqualini et al. 2005a) and perhaps from increased periods of larval wandering during host location (Hughes et al. 2003). In Bartlett pear, a high proportion of neonates enter via an opening in the calyx (especially early season when the fruit is hard) and pear ester may mask or interfere with this location behavior.

In conclusion, although pear ester seems amenable as a kairomonal adjuvant for use with insecticides, our data with CpGV in apple and pear are inconclusive. It is likely that practical improvements in formulation and application strategies (e.g., to optimize and maintain attractive release rates) and a better understanding of the role of environment (e.g., compare neonate host-searching behaviors in walnut and pome fruit) are needed.

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